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RESEARCH ARTICLE

Patterns of diversity in pigeonpea (*Cajanus cajan* (L.) Millsp.) germplasm collected from different elevations in Kenya

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Abstract Pigeonpea germplasm accessions collected from low (<500 m), medium (501–1000 m), high (1001–1500 m) and very high elevation zones (>1500 m) of Kenya were evaluated for 15 agronomic traits and seed protein content at ICRISAT, Patancheru, India. There were significant differences ($P < 0.001$) among elevation zones for the number of primary and secondary branches, days to 75% maturity, pod length, seeds per pod, 100-seed weight and seed yield. Mean values indicated that the accessions from low elevation zone were significantly different from those collected in higher elevation zones for early flowering and maturity, number of primary branches, pod length, number of pods per plant, seeds per pod, 100-seed weight, seed yield and harvest index. None of the accessions collected in Kenya belonged to extra early (<80 days to 50% flowering) and early (80–100 days to 50% flowering) maturity groups, as defined by time to flowering at Patancheru, India. Mean diversity index based on all characters indicated that accessions from the low elevation zone are more diverse than those from the higher elevation zones. Frequency distribution for trait extremes indicated that the accessions from the low elevation zone were early to flower and mature, short statured, produced more primary and secondary branches with high pod bearing length, long pods, more pods per plant, more seeds per pod, a high seed yield and harvest index. Accessions from the very high elevation zone were late flowering, with a large number of tertiary branches, large seeds and a high shelling percentage and could be a source for cold tolerance and the breeding of vegetable types. Results suggest that the elevation of collection sites is therefore a very important determinant of variation patterns of pigeonpea in Kenya.

Keywords Agronomic traits · *Cajanus cajan* · Collection sites · Diversity · Elevation · Pigeonpea

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Introduction

Pigeonpea (*Cajanus cajan* (L.) Millsp.) is an important legume of the tropics and subtropics. Because of multiple uses (source of food, fodder and fuel wood; material for thatching and fencing, and for soil improvement and windbreaks), it plays an important role in subsistence agriculture. Pigeonpea is grown as a field crop as well as backyard crop in more than 80 countries. During 2005, pigeonpea as a field crop was grown on an estimated 4.6 million ha, with a production of 3.5 million t and an average productivity of 0.90 t ha^{-1}. India has the largest area under pigeonpea (3.3 m ha) followed by Myanmar (0.58 m ha), Kenya (0.15 m ha), Malawi (0.12 m ha), Uganda (0.08 m ha), Tanzania (0.07 m ha), Nepal (0.03 m ha) and Dominican Republic (0.01 m ha) (FAO 2005). Since, in most pigeonpea growing countries pigeonpea is grown to a considerable extent in kitchen gardens, field bunds, in intercropped conditions, FAO estimates are considered to have not captured the entire extent of pigeonpea production.

The evolution of pigeonpea through natural and human selection in diverse elevation zones and under different cropping systems has resulted in a wide variety of locally adapted landraces. These landraces have evolved over years to fit into local cropping patterns and diverse end uses and represent a wide range of patterns of crop diversity. The knowledge of patterns of genetic variation of a crop species in any given region or country is very important for planning future germplasm collection missions and for efficient utilization of collected germplasm in crop improvement programs (Hayward and Breese 1993). Kenya, which lies between 4°S and 4°N on Eastern Africa's east coast is the most important pigeonpea growing country in Africa. More than half of the Kenya's land resources lie above 1,500 m.a.s.l. (Remanandan et al. 1982). The range of elevation of collection sites of accessions conserved in the ICRISAT genebank varies from 60 to 2000 m, within a narrow range of latitude, making temperature a major determinant of diversity patterns in Kenyan pigeonpea. This paper assesses the pattern of diversity distribution in relation to the elevation of pigeonpea germplasm collecting sites in Kenya.

Materials and methods

The R S Paroda genebank at ICRISAT, Patancheru, India has the global responsibility for collecting and conserving pigeonpea germplasm. Two germplasm collection missions were undertaken in Kenya in 1976 and 1982, which collected 63 and 247 pigeonpea samples, respectively (van der Maesen 1976; Remanandan et al. 1982). Collection was done over a wide range of elevation (60 m–2000 m.a.s.l.). Collection sites were classified as low (<500 m), medium (501–1000 m), high (1001–1500 m) and very high (>1500 m) elevations. This classification of the Kenyan collection resulted in 42 accessions (13.6%) from the low elevation zone, 68 (21.9%) from the medium elevation zone, 177 (57.1%) from the high elevation zone and 23 (7.4%) from very high elevation zone (Fig. 1).

Accessions were evaluated for 16 quantitative characters in different years at ICRISAT Center, Patancheru, A.P. India (17° 25′N latitude, 78° 00′E longitude and 545 m.a.s.l.) in vertisols during rainy season. Accessions were sown in three rows of 4 m long, with a spacing of 50 cm between plants and 75 cm between rows. The crop was fertilized with 20 kg N and 40 kg P_2O_5 ha^{-1} as basal dose, managed by recommended cultural and plant protection practices, including supplementary irrigation if required. Observations were recorded as mean of three representative plants from the middle row for plant height, number of primary, secondary and tertiary branches, number of racemes, pod bearing length, pod length, number of pods per plant, seeds per pod, 100-seed weight, shelling percentage, seed yield per plant and harvest index following the Descriptors for Pigeonpea (IBPGR and ICRISAT 1993). Observations on days to 50% flowering and days to 75% maturity were recorded on plot basis. Seed protein content was estimated with Technicon Autoanalyser (Singh and Jambunathan 1980). The data were analyzed using GENSTAT 6.1. The range, mean and variance for all traits were calculated for each of the four elevation zones. The differences between means of the different elevation zones were compared using the Newman–Keuls procedure for all traits (Newman 1939; Keuls 1952). The homogeneity of variances among the elevation zones was tested using Levene's test (Levene 1960), and phenotypic diversity was estimated by the Shannon and Weaver diversity index (H′) (Shannon and

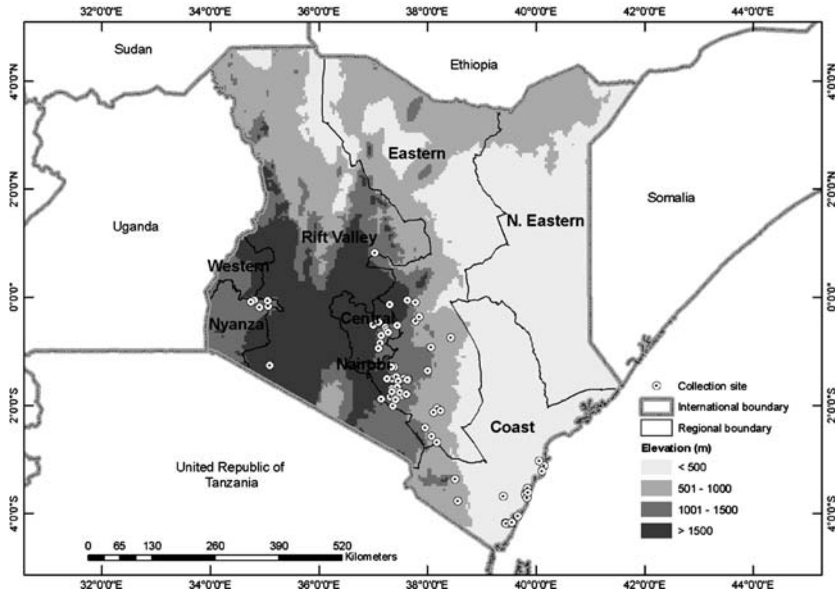

Fig. 1 Map showing pigeonpea germplasm collection sites at different elevations in Kenya

Weaver 1949). The index was estimated for each of the16 traits within each elevation zone. The frequency distribution of accessions for each extreme group within each character was calculated to identify the trait specific germplasm that is available at different elevations of Kenya.

Results and discussion

Kenya has a bimodal rainfall distribution with the short rains falling during October–November and the long rains during March–May (Remanandan et al. 1982). In Kenya, pigeonpea is usually sown during short rains, with major vegetative growth occuring during the long rains. Flowering commences after long rains followed by pod development during the cold dry months of June–July and the crop matures in the warm, dry months of August–September. Traditional types grown in Kenya are long duration (10 months), indeterminate types that are adapted to such a bimodal rainfall pattern (Remanandan et al. 1991). Because of varying climate, particularly, the minimum and maximum temperature differences in the four elevation zones, pigeonpea germplasm from Kenya is expected to vary considerably for many characters.

Variances among the elevation zones differed significantly ($P < 0.001$) for number of primary and secondary branches, days to 75% maturity, pod length, seeds per pod, 100-seed weight and seed yield (Table 1). Variances for days to 50% flowering, days to 75% maturity, plant height, pod length, seeds per pod, and seed yield per plant decreased with the increase in elevation of collection sites. Variances for number of secondary branches, number of racemes, pod bearing length and number of pods was highest in accessions from medium elevation zone.

The range and mean values for all characters are given in Table 2. The range among accessions was greatest in low elevation zone for number of primary branches, seeds per pod and seed yield per plant, in the medium elevation zone for days to 75% maturity, number of secondary branches, number of racemes, pod-bearing length, pod length and harvest index, and in the high elevation zone for days to 50% flowering, plant height, number of primary branches, number of tertiary branches, number of pods per plant, 100-seed weight, shelling percentage and seed protein content. Interestingly, the variation in the very high elevation zone was the lowest for all traits, except for seed protein content, which may be attributed to the natural selection and the intensive selection practiced by farmers at higher elevations.

The Newman–Keuls test of significance for mean values indicated that the accessions from

Table 1 Variances for various characters of pigeonpea germplasm collected from different elevations of Kenya

Character	Elevation (m.a.s.l.)					
	Low (<500)	Medium (500–1000)	High (1000–1500)	Very high (>1500)	*F* value[a]	Probability
Days to 50% flowering	561.81	353.77	329.67	228.51	1.86	0.1370
Days to 75% maturity	1034.04	380.04	337.86	131.08	8.59	<0.0001
Plant height (cm)	2451.92	1577.94	1581.33	1329.35	2.56	0.0550
Primary branches per plant (no.)	20.22	9.94	10.52	10.39	2.92	0.0340
Secondary branches per plant (no.)	84.74	96.49	48.45	72.06	2.69	0.0470
Tertiary branches per plant (no.)	12.46	20.72	31.89	15.17	0.32	0.8110
Racemes per plant (no.)	1478.13	2063.21	877.30	135.03	1.56	0.2000
Pod bearing length (cm)	118.12	276.77	170.22	17.95	2.26	0.0811
Pod length (cm)	2.07	1.10	0.79	0.74	6.70	0.0002
Pods per plant (no.)	6334.28	6977.28	4469.68	475.95	0.77	0.5111
Seeds per pod (no.)	0.72	0.30	0.25	0.14	8.70	<0.0001
100 seed weight (g)	3.77	7.00	10.67	7.53	3.39	0.0184
Shelling percentage	127.27	127.51	126.72	169.69	0.22	0.8830
Seed yield per plant (g)	1008.38	358.72	250.98	89.34	7.72	<0.0001
Harvest index (%)	33.49	38.01	28.24	10.96	1.05	0.3699
Seed protein content (%)	2.10	2.35	2.08	3.15	1.55	0.2031

[a] Variance homogeneity tested by Leven's test

low elevation differed significantly from those of other elevation zones for a number of traits (early flowering and maturity, primary branches number, pod length, number of pods, seeds per pod, seed yield and harvest index) (Table 2). Because of their early maturity, accessions from the low elevation produced pods with smaller seeds. As per the standard maturity classification followed at ICRISAT, none of the accessions from Kenya belong to extra early (<80 days to 50% flowering) and early (80–100 days to 50% flowering) maturity groups, when assessed at ICRISAT Patancheru. Only 12 accessions belong to the medium maturity group (100–130 days to 50% flowering) and 298 accessions to late maturity group (>130 days to 50% flowering) (ICRISAT 1978). Mean values for pod bearing length decreased with the increase in elevation, with accessions from the very high elevations having smallest pod bearing length (Table 2). Accessions from the low and medium elevations produced a significantly higher number of racemes than those from the high and very high elevations. Accessions from the medium, high and very high elevations were generally similar for number of primary branches, pod length, number of pods, seeds per pod, 100-seed weight and seed yield. The increased seed weight of pigeonpea accessions from higher elevations, where late maturing pigeonpeas are common, (Table 4) may be attributed to the prevailing low temperatures and consequently longer seed filling period similar to pearl millet (Ong and Monteith 1985). The elevation of origin has no significant effect on seed protein content.

Shannon–Weaver diversity index (H′) was estimated to compare phenotypic diversity among the accessions from different elevation zones based on all characters (Table 3). A low H′ value indicates low diversity and unbalanced frequency classes. The H′ estimates indicated considerable differences in diversity for all characters among accessions collected in different elevation zones. Mean diversity over all characters indicates relatively more diverse pigeonpea accessions in the low elevation zone than those from all other zones, which can be attributed to the prevailing optimum climatic conditions for pigeonpea growth in the low elevation zone. Averaged over the elevation zones, H′ values ranged from 0.42 ± 0.05 (number of pods) to 0.60 ± 0.01 (pod length). Diversity was greater in accessions from the low elevation zone for number of racemes, pod bearing length, number of pods per plant and harvest index; in the medium elevation zone for

Table 2 Range and mean values for different characters of pigeonpea germplasm collected from different elevations (in m.a.s.l.) of Kenya

Character	Range								Mean[a]			
	Low (<500 m)		Medium (500–1000 m)		High (1000–1500 m)		Very high (>1500 m)		Low (<500 m)	Medium (500–1000 m)	High (1000–1500 m)	Very high (>1500 m)
	Min	Max	Min	Max	Min	Max	Min	Max				
Days to 50% flowering	117	204	108	211	117	229	180	232	145.45d	174.69c	182.91b	191.17a
Days to 75% maturity	166	270	156	270	168	278	226	280	206.17c	242.46b	250.39ab	255.43a
Plant height (cm)	95	260	110	270	100	275	140	260	190.71a	202.28ab	210.25ab	219.57a
Primary branches per plant (no.)	2.3	21.3	2.3	18.7	2.0	21.0	3.3	16.7	10.85a	8.48b	7.84b	8.84b
Secondary branches per plant (no.)	4.7	48.0	2.7	49.7	3.7	48.7	2.7	33.0	19.81a	17.07ab	14.97b	17.88ab
Tertiary branches per plant (no.)	0.0	13.3	0.0	19.5	0.0	53.3	0.0	12.3	3.67a	4.21a	4.29a	4.99a
Racemes per plant (no.)	13.3	197.3	19.7	280.7	11.7	236.3	15.0	76.0	66.51a	57.56a	39.82b	36.82b
Pod bearing length (cm)	7.0	50.0	8.5	94.0	7.3	67.0	8.7	25.3	26.07a	27.3a	23.69a	17.2b
Pod length (cm)	5.8	11.6	4.3	10.5	5.5	10.1	5.4	9.2	8.35a	7.58b	7.7b	7.3b
Pods per plant (no.)	20.7	352.0	30.3	471.0	19.0	535.3	23.7	140.0	129.18a	98.7b	73.92b	66.56b
Seeds per pod (no.)	3.0	6.7	3.3	5.8	3.3	6.3	4.0	5.6	5.17a	4.64b	4.74b	4.73b
100 seed weight (g)	7.8	17.2	8.0	19.1	4.2	23.0	8.2	18.8	11.11b	14.21a	15.42a	15.15a
Shelling percentage	14.3	70.0	20.0	83.3	16.0	83.3	21.1	75.0	49.95a	53.2a	52.31a	54.53a
Seed yield per plant (g)	6.7	116.7	3.3	100.0	3.3	111.7	3.3	43.3	46.24a	26.03b	20.86b	19.85b
Harvest index (%)	2.6	28.5	1.7	34.7	1.7	32.2	3.6	15.2	13.66a	10.79b	8.65bc	7.72c
Seed protein content (%)	16.4	22.3	16.1	22.4	16.7	24.1	15.9	22.5	19.69a	19.82a	20.16a	20.25a

[a] Differences between means of different elevations was tested by Newman–Keuls test (same letter indicate non significant results)

Table 3 Shannon–Weaver diversity indices for various characters of pigeonpea germplasm collected from different elevations of Kenya

Character	Elevation (m.a.s.l.)				
	Low (<500)	Medium (500–1000)	High (1000–1500)	Very high (>1500)	Mean
Days to 50% flowering	0.49	0.61	0.62	0.36	0.52 ± 0.06
Days to 75% maturity	0.50	0.52	0.55	0.36	0.48 ± 0.04
Plant height (cm)	0.51	0.60	0.58	0.49	0.54 ± 0.03
Primary branches per plant (no.)	0.60	0.59	0.59	0.56	0.58 ± 0.01
Secondary branches per plant (no.)	0.56	0.55	0.58	0.56	0.56 ± 0.01
Tertiary branches per plant (no.)	0.46	0.46	0.37	0.51	0.45 ± 0.03
Racemes per plant (no.)	0.56	0.37	0.37	0.46	0.44 ± 0.05
Pod bearing length (cm)	0.56	0.44	0.50	0.56	0.52 ± 0.03
Pod length (cm)	0.57	0.60	0.62	0.61	0.60 ± 0.01
Pods per plant (no.)	0.54	0.34	0.34	0.46	0.42 ± 0.05
Seeds per pod (no.)	0.59	0.56	0.62	0.57	0.59 ± 0.01
100 seed weight (g)	0.55	0.60	0.63	0.52	0.58 ± 0.03
Shelling percentage	0.58	0.59	0.59	0.60	0.59 ± 0.00
Seed yield per plant (g)	0.50	0.47	0.45	0.54	0.49 ± 0.02
Harvest index (%)	0.59	0.56	0.52	0.42	0.53 ± 0.04
Seed protein content (%)	0.58	0.61	0.62	0.55	0.59 ± 0.02
Mean	0.55 ± 0.001	0.53 ± 0.020	0.53 ± 0.030	0.51 ± 0.020	

plant height; in the high elevation zone for days to 50% flowering, days to 75% maturity, pod length, seeds per pod, 100-seed weight and seed protein content; and in the very high elevation zone for pod bearing length, shelling percentage and seed yield per plant.

Considering the wide variation for different characters, accessions were grouped based on the extreme values of each character. For example, in case of days to 50% flowering, accessions that flowered in <130 days were considered as early and those flowered in >160 days as late flowering with reference to the Kenyan collection. This type of classification resulted in identification of the best sources of trait-specific germplasm for use in pigeonpea improvement. The low elevation zone was a more promising source of early flowering and early maturing accessions than all other elevation zones (Table 4). The frequency of early flowering and early maturing accessions decreased with the increase in elevation. None of the accessions from very high elevation zone were early flowering or maturing. Kenya lies on the equator, with day-lengths close to 12 h throughout the year (Remanandan et al. 1991). Pigeonpea is a quantitative short day plant, and most cultivars of all maturity groups flower readily in day-lengths of 11–11.50 h (Gooding 1962; Spence and Williams 1972). Thus no effect of photoperiod on flowering would be expected for pigeonpea in the country. However, the range of elevation in the country substantially modifies temperatures and consequently the rate of heat unit accumulation. Gupta et al. (1991) reported the relationships between latitude, altitude and temperature and observed a decrease of 1°C of mean annual temperature with the increase of 150 m in elevation. Therefore, pigeonpea germplasm adapted to high altitudes of Kenya could be a good source for cold tolerance. Averaged over 30 years, mean minimum temperature of all collection sites decreased from 22.8°C to 15.7°C and mean maximum temperature from 32.4°C to 27.9°C, with increasing elevation. Thus all but the low elevation zone have sub optimum temperatures for pigeonpea growth (Hijmans et al. 2002). Similarly, elevation, minimum and maximum temperature at ICRISAT, Patancheru location during the crop season is close to that of low elevational zone (Fig. 2). Whiteman et al. (1985) reported that flowering of all maturity groups occurs sooner in moderate temperature of 22–30°C, even under long days (12.5–16 h) and both low and high temperatures delay flowering in pigeonpea.

Table 4 Frequency distribution (%) for different modalities of various characters of pigeonpea germplasm collected from different elevations of Kenya

Character	Extreme group		Elevation (m.a.s.l.)			
			Low (<500)	Medium (500–1000)	High (1000–1500)	Very high (>1500)
Days to 50% flowering	Early	(<130)	23.81	1.47	0.56	0.00
	Late	(>160)	19.05	77.94	93.22	100.00
Days to 75% maturity	Early	(<180)	21.43	1.47	0.56	0.00
	Late	(>225)	28.57	82.35	92.09	100.00
Plant height (cm)	Short	(<130)	21.43	2.94	4.52	0.00
	Tall	(>225)	26.19	39.71	42.37	69.57
Primary branches per plant (no.)	Low	(<5)	9.52	11.76	20.34	8.70
	High	(>10)	50.00	26.47	19.21	26.09
Secondary branches per plant (no.)	Low	(<15)	35.71	45.59	56.50	39.13
	High	(>25)	23.81	19.12	8.47	17.39
Tertiary branches per plant (no.)	Low	(<5)	69.05	67.65	68.36	47.83
	High	(>10)	7.14	11.76	10.17	17.39
Racemes per plant (no.)	Low	(<30)	19.05	16.18	42.37	21.74
	High	(>150)	2.38	5.88	1.69	0.00
Pod bearing length (cm)	Short	(<20)	23.81	41.18	51.98	73.91
	Long	(>40)	14.29	14.71	11.30	0.00
Pod length (cm)	Small	(<6)	2.38	4.41	2.82	8.70
	Long	(>10)	11.90	1.47	0.56	0.00
Pods per plant (no.)	Low	(<100)	42.86	69.12	87.01	95.65
	High	(>250)	7.14	5.88	2.82	0.00
Seeds per pod (no.)	Low	(<4)	11.90	13.24	8.47	4.35
	High	(>5)	57.14	19.12	27.12	8.70
100 seed weight (g)	Small	(<10)	30.95	2.94	3.39	4.35
	Large	(>15)	4.76	38.24	53.67	65.22
Shelling percentage	Low	(<20)	2.38	1.47	1.13	0.00
	High	(>60)	16.67	23.53	19.77	43.48
Seed yield per plant (g)	Low	(<20)	30.95	51.47	70.06	60.87
	High	(>100)	7.14	0.00	0.56	0.00
Harvest index (%)	Low	(<10)	26.19	51.47	69.49	82.61
	High	(>20)	11.90	5.88	2.82	0.00
Protein content (%)	Low	(<18)	11.90	10.29	6.78	13.04
	High	(>20)	38.10	41.18	45.76	60.87

A high frequency of accessions (21.4%) from the low elevation zone was short (less than 130 cm), while 69.6% accessions from very high elevation zone grew tall (more than 225 cm) (Table 4). Generally, the frequency of short height accessions decreased and that of tall accessions increased with the elevation. None of the accessions from very high elevation zone were short. Plant height is related to maturity duration, photoperiod, temperature sensitivity and environment. Because of their prolonged vegetative growth, long duration pigeonpeas adapted to higher elevations grow tall (Remananadan 1990).

The low elevation zone was the best source of accessions with high number of primary and secondary branches, long pods, high number of pods per plant and seeds per pod, seed yield and harvest index. The medium elevation zone was the best source of accessions with a high raceme number and high pod bearing length. The very high elevation zone was the best source of accessions that produce more tertiary branches, large seeds, a high shelling percentage and a high seed protein content. Pigeonpea accessions from very high elevation are good sources for breeding vegetable types, because of their very large seed size.

The frequency of accessions with longer pod bearing branches, long pods, more pods per plant and more seeds per pod, a high seed yield and a high harvest index decreased with the increase in elevation, suggesting that the accessions from the

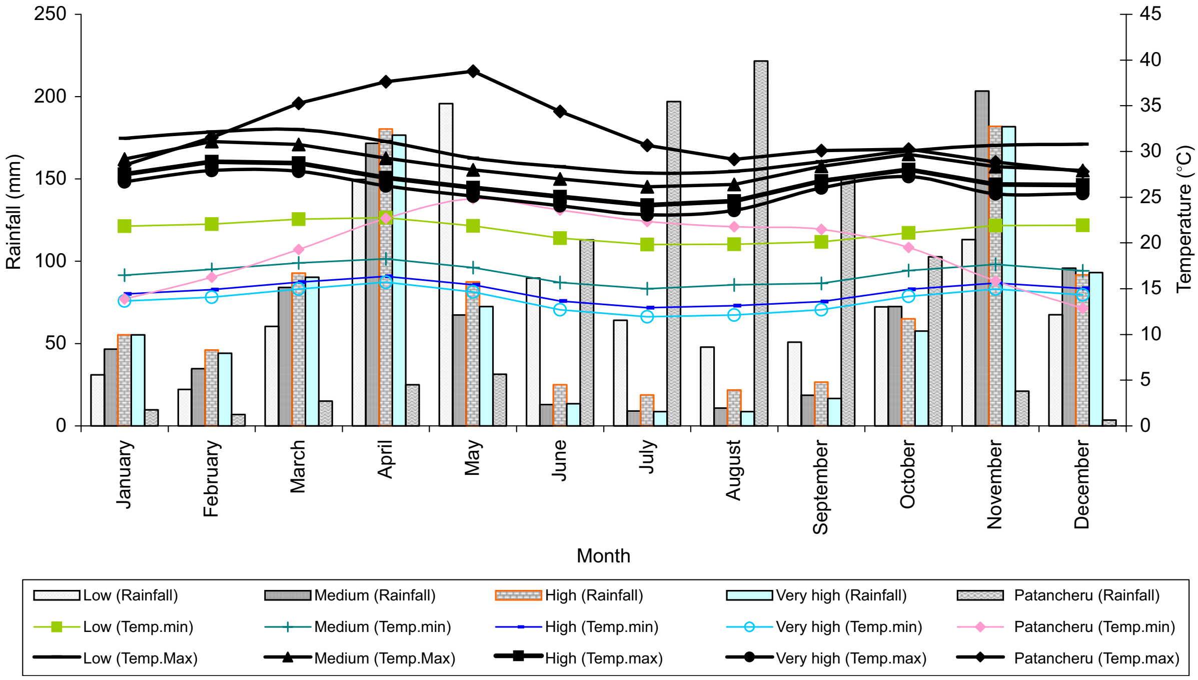

Fig. 2 Monthly mean minimum and maximum temperature and total rainfall at pigeonpea germplasm collection sites in Kenya and evaluation site at Patancheru, India

low elevation zone are the most promising for improving the yield components and yield. The differential behavior of accessions collected from varying elevations for different characters could be attributed largely to the temperature determined by the elevations (Gupta et al. 1991).

The results of our study imply that elevation of collection sites is an important determinant of variation patterns in a range of traits of pigeonpea in Kenya. Therefore, it is very essential to collect accurate and complete passport information, particularly on latitude, longitude, elevation, temperature and rainfall, to maximize the value of collected germplasm. This information will be very useful in predicting the genetic characteristics of the collected germplasm. Natural selection pressure for adaptation to different elevations, coupled with farmers' selection over many years at varying elevations also accounted for the observed diversity pattern. Though, the evaluation data used in the present study are preliminary in nature, these data reflect the differences in the genetic makeup of the accessions in terms of clear patterns of diversity. Further testing of trait specific germplasm at or near the place of origin will provide an opportunity to identify accessions with high potential for utilization in pigeonpea improvement.

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